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N-linked glycan characterization and profiling: Combining the power of a novel labeling reagent and a streamlined analytical platform for confident glycan assignments

Ying Qing Yu
Waters Corporation, USA

UPLC/FLR/MS (MS) analysis of released N-glycans labeled with a fluorescent tag has made routine with high performance LC and MS instrumentations. Glycans labeled with conventional fluorescent tags such as 2-AB and 2-AA can be detected by Fluorescent (FLR) detector with ultra-high sensitivity. Unlike FLR detector, mass spectrometry is known to be less sensitive to detect native or tagged glycans especially the low abundant ones due to their poor ESI performance. The limited dynamic range of this approach has been restricting the utilization of this combined automated workflow for glycan characterization. In order to overcome the low MS ionization efficiency associated with the conventional labels and confidently assign lower level glycans, a novel tag, RapiFluor-MS™ (RFMS) developed by Waters Corporation has been employed. RFMS contains a rapid tagging reactive group, an efficient fluorophore and a functional group that imparts high ionization efficiency. Complete tagging of glycans can be achieved in less than 5 minutes using this novel reagent. Initial results with the new glycan label show significant enhancement in both the FLR and MS (MS) signals compared to 2-AB. The increased sensitivity enables the detection and identification of very low level glycans at (0.1%) with sufficient MS signals. In this study, we demonstrate the benefits of combining the novel labeling reagent RFMS with an integrated UPLC/FLR/Xevo G2-XS QTOF MS system for detailed characterization of the minor glycoforms from therapeutical proteins.

Biography

Ying Qing Yu is a Principal Scientist in the Pharmaceutical group at Waters Corporation. She joined Waters Corporation since 2001, shortly after she received her PhD from the Analytical Chemistry Department at Purdue University. She is a group leader in the Biopharmaceutical lab unit at Waters. Her group's focus is on Protein Biotherapeutics characterization using UPLC/QTOF MS platform. The projects she is currently working on range from glycan profiling, peptide mapping and lately the Hydrogen Deuterium Exchange Mass Spectrometry for protein higher order structure analysis. She has extensive experience in Mass Spectrometry, Gas-Phase Ion Chemistry and Liquid Chromatography Separation Techniques.

Ying_Qing_Yu@waters.com

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